UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

EPA-SAB-EHC-92-001

October 10, 1991

OFFICE OF
THE ADMINISTRATOR

Honorable William K. Reilly
Administrator
U. S. Environmental Protection Agency
401 M Street, S.W.
Washington, D.C. 20460

Subject: Review of the Office of Research and Development's Draft Strategy for Health Effects Research on Chemical Mixtures.

Dear Mr. Reilly:

The Science Advisory Board's Environmental Health Committee met February 28 and March 1, 1991 in Dallas, Texas to review the Office of Research and Development's (ORD) Draft Strategy for Health Effects Research on Chemical Mixtures.

The charge to the Committee incorporated the following questions:

a. Has the strategy generally captured the appropriate issues?

b. Is the strategy focusing on the appropriate issues?

c. Given the overarching issues, is the scope of activities identified about right?

d. Is the mixtures research strategy consistent with recommendations in the Science Advisory Board's (SAB) reports *Future Risk* (1988), and the more recent *Reducing Risk* (1990)?

e. Does the Fiscal Year 1993 Cross Media Research Initiative appear to reflect an appropriately balanced approach to implementing the strategy?

Exposure to chemical mixtures (rather than to single chemicals) is a common problem for EPA. In the absence of definitive information about the mixture itself, the Agency adopted interim procedures for calculating potential health risks posed by mixtures (51 FR 34042-34054, September 24, 1986). The procedure assumes that the total risk of a mixture is equivalent to the sum of the risks associated with the mixture's constituents.

The scientific cogency of such an approach, in the absence of knowledge that each constituent acts independently, is debatable. Risk estimates based on simple additivity may either overestimate or underestimate the true risk. Current data indicate that, under some circumstances, combinations of chemicals may be more toxic than would be predicted by summing the toxicity of the individual chemicals themselves; in other cases, however, the combination could be less toxic than the sum of the individual toxicities.

To help narrow the gaps in knowledge about the health risks of mixtures, the ORD proposed a research strategy based on a bottom-up approach--a strategy which presumes that once the constituents of a mixture have been identified, and the individual dose-response functions elaborated, the toxicity of the total blend can be calculated--provided that the mechanisms governing interactions are understood.

The Committee agrees that complex mixtures present a significant public health issue. We are not convinced, however, that ORD has devised the most useful research program for this purpose. Specifically, we have concerns on the following three issues:

a. Is the emphasis on "bottom-up" testing feasible? Many mixtures include dozens to thousands of constituents. The Committee is pessimistic about the feasibility of predicting the total potency (or the potency of the major constituents) of such a mixture by some combinatorial algorithm?

b. How does the proposed research program plan to examine the variable of dose? We are disturbed to note that the proposed strategy document favors classical high-dose testing of single compounds as the components of an additive model. Most current experiments on mixtures involve high-dose studies of binary combinations, but EPA's risk analyses must address lower, environmental doses.

c. What is the nature of the research to be carried out? The Committee found the strategy to be somewhat vague in its descriptions of specific research initiatives, and unclear about the resources required to support such a program. In its present form, the document lacks a depth of discussion and exposition sufficiently compelling to warrant full support. This is not to imply that such a case cannot be made--only that it presently is lacking. The draft strategy contains much useful information however, and once the problems of focus and logic noted in the EHC report are addressed, has the promise of being a truly useful strategic document.

The Committee recommends that:

a. Before undertaking a significant expansion of its efforts on the health risks of complex mixtures, ORD should frame more specific plans for what it hopes to accomplish. It needs to provide a clearer picture of its priorities and a system for generating them. Existing, rather than hypothetical scenarios should guide the direction of the research. The highest priorities should be given to those activities which will improve the Agency's ability to assess the public health risks of complex mixtures. Specifically, methods to assess these risks must be improved and validated, and information must be developed to facilitate the assessment of specific complex mixtures.

b. ORD should consider their approach to a mixtures research strategy within the context of the Agency's adoption of the Risk Reduction program. Are major

investments in complex mixture questions warranted without further examination of the benefits they might yield? Current evidence of interactions that inflate toxicity is based on high dose scenarios. Equivalent phenomena may not appear at low environmental exposures (as is suggested by the 1988 National Academy of Science report, *Complex Mixtures: Methods for In-vivo Toxicity Testing*), the region of the dose-response function that requires the most research emphasis.

Lastly, the SAB Executive Committee, in its review, raised one issue not addressed in the EHC report. A statement in the ORD draft strategy document (top of page 14) implies that the commonly accepted NAS risk assessment paradigm is not satisfactory for dealing with issues involving complex mixtures. We do not agree, and in fact believe that the paradigm is most useful when dealing with the complexities and uncertainties posed by problems such as complex mixtures

In summary, the document is a useful attempt to integrate work by other government agencies and the broader scientific community. Its strength comes from its approach to the design of a research plan which, although vague and perhaps unrealistic, is nevertheless a prescription rather than another diagnosis. It offers premises that can be argued. Its weakness is a reliance on default assumptions that few would agree are scientifically defensible.

We appreciate the opportunity to review this proposal, and look forward to receiving your response to the issues we have identified.

Sincerely,

Raymond C. Loehr

Dr. Raymond C. Loehr
Chairman
Science Advisory Board

Bernard Weiss

Dr. Bernard Weiss
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Environmental Health Committee

ENCLOSURE

United States
Environmental
Protection Agency

Science Advisory
Board (A-101)

EPA-SAB-EHC-92-001
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EPA

A RESEARCH STRATEGY FOR ASSESSING THE HEALTH EFFECTS OF EXPOSURE TO COMPLEX MIXTURES

REVIEW OF THE OFFICE OF RESEARCH AND DEVELOPMENT'S DRAFT STRATEGY FOR HEALTH EFFECTS RESEARCH ON EXPOSURE TO COMPLEX MIXTURES BY THE ENVIRONMENTAL HEALTH COMMITTEE

U. S. ENVIRONMENTAL PROTECTION AGENCY

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ABSTRACT

The Environmental Health Committee (EHC) of the EPA Science Advisory Board (SAB) was asked to review a strategy document, developed by the Agency's Office of Research and Development (ORD), for conducting research on the health risks of exposure to complex chemical mixtures. Such mixtures are not only pervasive in the environment, but also represent the dominant mode of chemical exposure for the U.S. population. The Committee met on March 1, 1991 in Dallas, Texas to receive briefings from Agency officials and discuss the following issues: a) Has the strategy generally captured the appropriate issues, both technically, and in terms of the mission of the EPA?; b) Is the scope of activities correct?; c) Is the research strategy consistent with recommendations in the SAB's *Future Risk* and *Reducing Risk* reports?; d) Does the Fiscal Year 1993 ORD Cross Media Research Initiative reflect a balanced and sufficiently substantial approach to implementing the strategy?

The Committee found that a basis for a major expansion of current efforts is not cogently presented in the document. The coupling between the research program, which is not described in specifics, and the Agency's thrust toward risk reduction, remains vague.

The EHC views validation and improvement of the methods applied to the risk estimation of mixtures as a primary objective of ORD programs in this area. Current translations into risk assessment and regulatory decisions rely upon the additive model (the assumption that algebraic summation of dose is the most reasonable default position). The proposed research expansion emphasizes what is called the bottom-up approach, defined as the identification of mixture components, followed by a study of their joint actions and how these might be modified by various biological mechanisms. A relative ranking of the priorities for such a program, or, at least, the means by which priorities will be established, needs to be devised by ORD.

The Committee also sees tests of interactions at low doses as a top priority, recognizing that such tests may yet have to be developed. Studies of interactions require exploration of the entire dose-response function. If such research then fails to detect a significant problem at these low exposure levels, the issue of inflated toxicity due to interactions might be assigned a lower ranking in EPA's list of priorities for risk reduction. Instead, more efficient techniques for determining the comparative potencies of truly complex mixtures should receive greater emphasis. The comparative costs and time requirements of bottom-up and top-down approaches, including bioassay-directed fractionation for the latter, should be calculated for each mixture to be tested.

Complex mixture issues transcend EPA's purview and also involve the Food and Drug Administration, the Department of Energy, the Department of Agriculture, the National Institutes of Health, and others. Generic problems should be shared with the other agencies.

Keywords: Complex mixtures; additive toxicity; dose-response ranges; interaction; reference dose.

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1. EXECUTIVE SUMMARY

The EPA's Office of Health Research (OHR), and its associated Health Effects Research Laboratory (HERL), have compiled a strategy document designed to guide future research on the health risks of complex chemical mixtures. Such mixtures are not only pervasive in the environment, but also represent the dominant mode of chemical exposure for the U.S. population. The Environmental Health Committee (EHC) of the Science Advisory Board (SAB) was asked to review the strategy document. The Committee's comments and recommendations are presented under the following headings: Significance of The Area, Priorities, Implications for Risk Assessment, Allocation of Resources, Scientific Benefits, Document Structure, and Appropriateness

The Committee recognizes that all risk estimates ultimately are framed, whether intentionally or unwittingly, in a multiple chemical context; even in laboratory research, diet may modify the response to chemicals under examination for toxicity. The basis for a major expansion of current efforts, however, is not cogently presented in the document. The coupling between the research program, which is not described in specifics, and the Agency's thrust toward risk reduction, remains vague. In its present form, the draft strategy lacks a depth of discussion and exposition sufficiently compelling to warrant full support. This is not to imply that such a case cannot be made--only that it presently is lacking.

The EHC views validation and improvement of the methods applied to the risk estimation of mixtures as a primary objective of OHR programs in this area. Current translations into risk assessment and regulatory decisions rely upon the additive model; that is, the assumption that algebraic summation of dose (as a proportion of the RfD), to calculate a Hazard Index, is the most reasonable default position. The proposed research expansion emphasizes what is called the bottom-up approach, defined as the identification of mixture components, followed by a study of their joint actions and how these might be modified by various biological mechanisms. A relative ranking of the priorities for such a program, or, at least, the means by which priorities will be established, needs to be devised by OHR.

Although a substantial literature on combinations is available, it suffers from two shortcomings. First, it is dominated by mixtures of only two components, in contrast to most complex mixtures in the environment, which often consist of thousands of chemicals, many of them not even identified. Second, it almost exclusively is confined to interactions at high doses, in contrast to the situations prevailing in environmental exposures. Nonetheless, we were disturbed to note that the proposed strategy document favors classical high-dose testing of single compounds as the components of an additive model.

For these reasons, the Committee views tests of interactions at low doses as the earliest priority, recognizing that such tests may yet have to be developed. That is, any studies of interactions, including mechanistic studies, require exploration of the entire dose-response function. If such research fails to detect a significant problem at these low exposure levels, as suggested by dose-response modeling by Committee members and by the National Academy of Science (NAS) (1988) report, then the issue of inflated toxicity due to interactions can be assigned a lower ranking in EPA's list of priorities for risk reduction.

Instead, more efficient techniques for determining the comparative potencies of truly complex mixtures, as defined in the NAS report, should receive greater emphasis, especially as they contribute to the Agency's regulatory and informational requirements. The comparative costs and time requirements of bottom-up and top-down approaches, including bioassay-directed fractionation for the latter, should be calculated for each mixture to be tested.

The Committee also noted that complex mixture issues transcend EPA's purview and also involve the Food and Drug Administration, the Department of Energy, the Department of Agriculture, the National Institutes of Health, and others. Generic problems, such as appropriate modeling, should be shared with the other agencies because, by current methods, even modeling the joint actions of two chemicals presents mathematical challenges, as noted in the National Academy of Science (1988) report.

If the strategy document is scheduled for revision, the EHC recommends an expansion of how the research program is to be implemented, how it derives from EPA's broad responsibilities, and how it relates to other HERL programs. It should clarify its relation to and impact upon the Hazardous Waste and Superfund programs, Pesticides and Toxic Substances programs, Drinking Water programs, and other initiatives.

In summary, the document is a useful attempt to integrate work by other government agencies and the broader scientific community. Its strength comes from its approach to the design of a research plan which, although vague and perhaps unrealistic, is nevertheless a prescription rather than another diagnosis. It offers premises that can be argued. Its weakness is a reliance on default assumptions that few would agree are scientifically defensible.

2. INTRODUCTION

2.1 Background

The EPA's Office of Health Research (OHR) and its associated Health Effects Research Laboratory (HERL) have recognized the need to perform research which will provide the Agency with a better understanding of the potential health risks posed by exposure to chemical mixtures. OHR also recognized that, in order to maximize the impacts of its efforts, the chemical mixtures research program it undertakes must have well defined goals and a strategy to achieve them. OHR is currently developing a research strategy to guide its chemical mixtures program over the next five to ten years. To ensure adequate consideration of the decisions it must make and of the scientific directions required, OHR requested a review of its strategy document by the SAB. The EHC received briefings on the subject by Agency officials at its meeting of March 1, 1991, in Dallas, Texas, and discussed the issues noted below. The report which follows stems from those discussions.

2.2 Charge to The Committee

The charge to the Committee for this review posed the following questions:

a. Has the strategy generally captured the appropriate issues? Are there any major areas that have been overlooked and which should be included?

b. Is the strategy focusing on the appropriate issues for attention by the United States Environmental Protection Agency's (EPA) Office of Research and Development's Health Effects Research Laboratory (HERL).

c. Given the overarching issues, is the scope of activities identified for ORD/HERL involvement about right?

d. Is the mixtures research strategy consistent with recommendations in the Science Advisory Board's (SAB) report Future Risk (1988), and the more recent Reducing Risk (1990), as well as ORD's Long-Term Research Program?

e. Does the Fiscal Year 1993 Cross Media Research Initiative appear to reflect an appropriately balanced and sufficiently substantial approach to implementing the strategy?

3. COMMENTS AND RECOMMENDATIONS

The recommendations of the Committee are organized in the following areas: Significance of The Area, Priorities, Implications for Risk Assessment, Allocation of Resources, Scientific Benefits, Document Structure, and Appropriateness.

3.1 Significance of the Mixtures Research Area

3.1.1 Are Mixtures a Significant Enough Problem to Warrant a Major Expansion of Effort?

Exposure of humans to complex mixtures poses a challenge in risk estimation and risk reduction. For example:

a. Ambient air pollution is of concern for metropolitan areas subject to certain meteorological conditions which combine exposures to ozone, nitrogen oxides, sulfur dioxide, particulates and hydrocarbons. The EPA has submitted a draft document characterizing the risk of excess cancer resulting from exposure to 64 air pollutants. The 64 toxicants were quantified in urban areas and represent products of incomplete combustion and organic solvents, including many halogenated hydrocarbons. Risk estimates were based on the assumption of additivity. Sensitive populations include the very young and those who are compromised by respiratory and cardiac diseases.

b. Drinking water in certain areas has been associated with excess cancer levels. Disinfectant by-products of water purification chemicals are the most likely suspects.

c. The large number of hazardous waste sites and their remediation have driven the EPA's need to characterize the risk of exposure to complex mixtures.

3.1.2 Is the Program Broad Enough?

The program may, in fact, be excessively broad; it spans most of toxicology. The research strategy should be better focused to provide data appropriate for the EPA mission of environmental risk reduction (usually achieved through regulation). The EPA is a regulatory agency which serves and communicates with Congress, state and local governments, environmental and business groups, specific industries, and the public at large. The mission of the research arm of the EPA must be to supply data on which to base regulatory activity.

3.2 What is the Highest Priority for Such a Program? Conversely, Which Aspects Are Less Compelling and Less Prominent in Risk Reduction?

The highest priorities should be given to those activities which will improve the Agency's ability to assess the public health risks of complex mixtures. Specifically,

methods to assess these risks must be improved and validated, and information must be developed to facilitate the assessment of specific complex mixtures.

The Environmental Health Committee recommends that, initially, higher priority should be given to the improvement of risk assessment methodologies, because its resolution generally precedes risk evaluation of most of the complex mixtures in the Agency's domain.

In satisfying this goal, there are at least three generic issues that merit emphasis:

a. Currently, the Agency assumes additivity in the absence of specific information for a mixture. This is reasonable for a wide variety of mechanisms and assumptions about interactions; however, it is important to identify those mechanisms and outcomes for which additivity may be less appropriate. Some examples of non-additive interactions are given on pages 2-18/19 of the strategy (these include, among others, interactions due to absorption, accelerated metabolism, inhibition of metabolism, alteration of renal excretion and direct chemical or physical interactions). Cases in which the additivity assumption may not be correct can be identified by assessing the compatibility of the additivity assumption with what is known about the mechanisms associated with various endpoints, as well as with underlying biochemical processes. What the proposal means by "mechanisms" is ambiguous, however, and must be clarified before undertaking an experimental program, particularly in the context of exposure/dose levels. The Committee therefore recommends a sharper focus on, and clearer definition of, mechanisms. It is especially critical to evaluate the role of dose level in addressing the reasonableness of the additivity assumptions (see Recommendation (a)).

b. Dependence upon the top-down approach is hampered by the large number of potential mixtures, which are highly variable. The Committee recommends an effort to compare the potencies of mixtures of the same composition and how the potency of such mixtures may vary with changes in relative quantitative composition. A single assay may not provide sufficient results to estimate absolute potency, but it can yield estimates of relative potency. The appropriate assays and measures suitable for comparative potency approaches need to be identified; i.e., how do we relate (through which measure) the potency of one mixture to another? Selected biomarkers could be one important component of this effort. Another important consideration is the relationship between short-term and long-term toxicity measures.

c. More emphasis should be given to the significance of "complex" in complex mixtures. Many, perhaps most, complex mixtures cannot be characterized fully. Some contain thousands of individual components. Additivity is applied only to those constituents which can be identified, and for which toxicity data are available. The impact of less than complete complex characterization needs to be assessed by a process such as fractionation (cf, NAS, 1989). Simple ways to assess adequacy of characterization could be defined. These could include some of the same methods examined/developed above.

For generic issues, the Committee recommends reduced emphasis on structure-activity relationships and pharmacokinetics in the context of complex mixtures. These areas,

although important, are not yet fully developed/applied for individual compounds. Their application to simple mixtures and, especially, to complex mixtures represent much later stages.

Complex mixtures of particular concern, because of wide-spread prevalence, for example, should generate their own research agendas. The list on page 2-27 of the strategy document includes many of these substances (among which are PCBs, dioxins, furans, VOCs, PAHs, metals and products of incomplete combustion). Priorities for these mixtures should relate to regulatory needs and informational gaps. EPA personnel should be equipped to determine these priorities.

3.3 How Would a Mixtures Program Contribute to Reducing Uncertainty in Risk Estimates Apart From the Generic Questions, Such as Dose-response Modeling, that it Embodies?

3.3.1 Recommendation 1.

The Committee recommends that the Agency investigate circumstances under which chemicals that produce adverse effects by different mechanisms are likely to produce deviations from additive or multiplicative risks at the low doses or exposure levels usually prevailing in the environment.

The current default assumption for estimating the risk of a mixture calculates the total risk for an endpoint by summing the risks of the individual components. This may be a reasonable assumption for chemicals operating by different mechanisms at low doses in the absence of saturation and competition for receptors. Even if the relative risks are multiplicative, the combined risk will still be approximately additive at low doses (e.g., if relative risks of 1.01 and 1.02 for two components are multiplicative, the total relative risk is 1.01 x 1.02 = 1.03). Although the question must be posed about which circumstances and with what frequency more than multiplicative risks occur, the likelihood of such a possibility seems low.

3.3.2 Recommendation 2

For chemicals that produce an adverse effect by the same assumed mechanism, which is equivalent to increasing the dose of a single chemical, the Committee recommends that the Agency conduct studies to identify and measure the active chemicals, or their metabolites, at the target tissue site to confirm that the dose additivity model predicts the magnitude of the adverse effect.

For example, all of the individual components in a mixture may be below an assumed threshold dose, but the sum of their doses may exceed a threshold dose, thereby producing a detectable effect. The toxicity of many classes of compounds may be additive, for example non-carcinogenic polycyclic aromatic hydrocarbons, aliphatic solvents, halogenated organic solvents, organophosphates, and halogenated pesticides. If the endpoint is cancer and a chemical produces a carcinogenic metabolite in the detoxification process, any substance that increases the half life of the carcinogenic moiety will increase the incidence of cancer.

3.3.3 Recommendation 3.

The Committee recommends that the Agency explore circumstances under which chemicals can alter the mechanisms of toxicity, either qualitatively or quantitatively, of other chemicals when they occur together.

A question to be addressed by a mixture research program is whether certain chemicals or classes of chemicals may mutually alter, either qualitatively of quantitatively, their actions at low doses. Risk or dose additivity cannot be assumed under these circumstances. Such explorations may help reduce the uncertainty of risks based solely on the default assumption of additivity.

3.4 Does the Proposed Strategy Represent A Proper Allocation of EPA Resources According to the Proposed Budget?

In general, the present draft document places major emphasis on a "bottom-up" testing program designed to test the assumed additivity of a biologic response to individual mixture components. This strategy would require a very large commitment of funds as well as significant manpower over a long period of time. Relevant concerns involve several important, but presently unanswered, questions. For instance, would not the "top-down" method (starting with the mixture) be of equal scientific utility? Would it require a somewhat lower funding level and less time? It is recommended that this question be resolved, perhaps by a comparative cost analysis. This issue might also be resolved by an analysis of the available data, which could provide indications and documentation that one means appears to be more useful than the other.

Another topic missing from the present draft document is a projection of the anticipated enhancement of public health provided by either testing mode. Decisions about funding require a careful weighing of priorities comparing this topic to other areas of public health concern. The program could enhance (or reduce) its funding priority and general acceptance by the scientific community if such documented comparisons were added to the report. It is recommended that this be provided.

In its present form, the report lacks a depth of discussion and exposition sufficiently compelling to warrant full support. This is not to imply that such a case cannot be made--only that it presently is lacking.

The Committee recommends that the means by which priorities for testing, and priorities for taking on specific types of toxicologic tests, be specified. We also recommend that a decision tree or other means be developed for prioritization of toxicological tests. This is particularly relevant in developmental (although presently less so for reproductive) toxicology where several in vitro (or even short-term in vivo) assays could readily assist in establishing priorities for higher level tests. An option for the Agency would be to establish priorities for remediation by applying bottom-up assay techniques (a strategy which presumes that once the constituents of a mixture have been identified, and the individual dose-response functions elaborated, the toxicity of the total blend can be calculated--provided

that the mechanisms governing interactions are understood), then selecting remediation procedures by top-down procedures (that is, addressing the total toxicity of a given mixture).

3.5 Could the Strategy Be Modified to Enhance its Contribution to Toxicological Principles?

3.5.1 Recommendations

a. The EPA should seek to involve other agencies with major experience in health-related research as it expands its research on mixtures. For instance, the Food and Drug Administration (FDA) has to contend with combinations of food additives.

b. The EPA should acknowledge its limited scope of interests (reducing enviromental risk) within the broader universe of mixtures-related issues. The Agency should determine how its limitations determine approach to specific mixtures. It must then develop a strategy for assigning priorities.

c. The Agency should emphasize research on the generic questions involved in dealing with mixtures. These include short-term tests to evaluate the validity of the default (additivity) model and how it varies with dose.

3.5.2 Findings

Humans are exposed to complex mixtures in the form of food, drugs and xenobiotics. Single agents of concern are always one component of a complex background, and the composition of this complex mixture is variable. The health experiences of humans are known to be affected by this "chemical soup," and there are many examples of interactions between xenobiotics, drugs and foods.

At this time the EPA is unable to investigate the totality of this mixture problem, and, in fact, is examining only a small subset of the whole mixture, namely the xenobiotics. This leads to a poorly specified problem statement, because it is assumed that only the exposures to the xenobiotics produce adverse effects, and that there will be no interactions with foods or drugs.

Issues related to the toxicity of complex mixtures transcend the specific interests of the EPA and include the mandates of other organizations with concerns in the health sciences, especially the FDA, the Department of Energy (DOE) the Department of Agriculture, the National Institutes of Health (NIH), the National Academy of Science, and others.

Problem areas with potentially universal applicability should receive high priority for investigation. Examples of such problem areas are:

a. Generic characteristics of the dose-response relationship of mixtures; for example do they simulate the dose-response parameters of individual components? Do they show unusual properties at low doses?

b. Further development of short-term tests that are sufficiently sensitive to describe low-dose parameters of the dose-response function and that are suitable for bioassay-directed fractionation.

Historically, research on the generic properties of truly complex mixtures has received little support; the predominant questions have driven research on binary mixtures, usually in the form of plotting isobolograms or full dose-response functions for each component. Such an approach is not feasible for complex mixtures; the number of combinations for only a few components is staggering.

Even modeling the joint actions of two chemicals, to derive dose-response surfaces, presents mathematical challenges (NAS, 1988). The challenges expand exponentionally beyond binary mixtures, even if one assumes linear dose-response relationships, an absence of thresholds, and similar effects on the target or receptor. If there are thresholds for some components of the mixture and/or there are non-linear dose-response curves, predictive models would be expected to fail on mathematical grounds alone.

3.6 The Structure of The Document And Its Modification

3.6.1 Organization of The Report

The report does an effective job of outlining the major policy issues associated with complex mixtures. There are however, problems with the description of the proposed research in section 4, which is divided into parts dealing with hazard identification research, exposure assessment research, dose-response assessment research, mixture-specific testing and cross-cutting testing. The HERL has chosen to focus on the bottom-up strategy with emphasis on interactions and, particularly, the issue of additivity. The rationale which leads to this circumscribed focus is not effectively presented. As an example, the second paragraph on page 4-I indicates that "the most desirable data" for risk assessments are obtained from direct test of the mixtures. The authors indicate that these top-down data are not generally available and use this deficiency as an indirect justification for a bottom-up approach. If these data are not available but are most desirable, why not design a program to obtain such data? The introduction to the proposed research strategy should offer a positive, direct justification for the proposed strategy.

A more basic problem is the terse discussion of the actual research strategy proposed by HERL, as presented in section 4.3. The discussion indicates that the program will focus on issues that will improve bottom-up dose-response assessments (e.g. improve predictability). The major components include an evaluation of the additivity assumption, the development of quantitative models, and the effects of mixtures on uptake/distribution of chemicals. Although these seem to be reasonable aims, the specific strategies to guide these efforts remain unclear. Mechanisms of interaction are mentioned briefly in section 4.3.1 and several questions are presented in section 4.3.3, but these discussions are too superficial to

allow a meaningful evaluation of the proposed research or its relationship to other programs within or beyond EPA purview.

3.6.2 Recommendations

a. The introduction to section 4 should be rewritten to provide a stronger and clearer justification for the proposed research strategy.

b. The proposed strategy should be elaborated upon so that the major issues to be addressed can be more clearly understood.

c. The relationship of the proposed research to other programs within the Agency could be more clearly delineated. This would be facilitated by recommendation (b).

d. Section (4) should be reorganized so that the major thrusts of the strategy are given in the introductory section (i.e. not 4.3).

3.7 Does the Research Initiative Include Appropriate Topics, Approaches and Priori ties?

The adequacy of the proposed 1993 initiative is a difficult question to address. The basic reason for this difficulty is that, while many of the research areas are certainly of importance to the individual offices within the Agency, there does not appear to be any cohesive force which will allow the Agency to evaluate their progress in dealing with the whole mixtures question.

The term "research focus" is used throughout the document. As noted, however, the focus is generally on specific questions and specific mixtures, not on comprehensive strategy. This is unfortunate because the overall goal of validating the additivity assumption is only a beginning model for experimental design. The Committee recommends that the Agency more carefully review how the suggested projects will contribute to fulfilling the EPA's responsibilities.

Although the Committee recognizes the financial constraints under which HERL operates, and its dependence on support from the regulatory program offices for specific projects, it strongly recommends that a more global view be adopted. Additional financial support can be negotiated for studies on the mixture problem because it is of importance to virtually all of EPA's regulatory programs. This is not only logical from a scientific perspective but is made in accordance with the EPA document, Reducing Risk, which states the Agency's position that long term goals such as these should not be driven by existing programmatic structures or specific current regulatory requirements.

At HERL, the research should extend beyond merely augmenting a small existing program dealing with evaluating whole and fractionated complex mixtures. The Committee recommends the program go well beyond the genotoxicity endpoints by adding many other

endpoints currently at its disposal. It also should put methodological issues and development near the top of its concerns and not relegate these important areas to only a reactive position.

The work proposed in the Air Program probably represents one of the Agency's best approaches so far to the evaluation of truly relevant and realistic mixtures. The Committee recommends that the Agency continue to support fully this effort and to add new endpoints as described. Careful evaluation of these studies, including species comparisons, is likely to add interpretable data, especially in the context of the validity of the additivity model.

The Hazardous Waste and Superfund research focus on metals and PCBs does not clearly delineate how these studies will test the additivity hypothesis. Since they are ubiquitous components of hazardous waste sites, these chemicals are of extreme importance, and it would seem logical that these pertinent studies be undertaken. The Committee had difficulty in determining the relevance of these studies to the more general picture of testing additivity or other models.

The Pesticide and Toxic Substances research likewise has the reasonable aim of investigating the interaction of "inert" and "active" ingredients utilized in existing pesticide formulations. However, the Committee recommends a broader evaluation than the one stated, and that prior to such studies, a more detailed consideration be given to how the data will be interpreted if the studies are undertaken; for example, how will low dose extrapolation be addressed?

The Drinking Water research focus is also blurred. Although the Committee recognizes that developmental and immunotoxic endpoints are important, up until now these have seldom provided the scientific endpoints that determine the regulatory standards, namely, maximum contaminant level goals and maximum contaminant levels. The Committee recommends that the Agency review these data in undertaking to determine the validity of the additive model. That is, the already established criteria and endpoints of toxicity should precede those proposed for inclusion in deriving meaningful numbers for mixtures.

The multimedia research focus outline presents interesting concepts that may indeed be relevant, e.g., the impact of one chemical on the pharmacokinetics and ultimately toxicological properties of another. However, the Committee recommends that much more definitive plans, based on real scenarios be carefully outlined before specific studies are undertaken.

3.8 Has This Document Made Adequate Use of Previous Efforts, Such as The NAS/National Research Council Report (NRC) ?

The authors of the document have made extensive use of previous efforts, especially the NAS "Complex Mixtures" report, and they have expanded on this document (and others) by adding the survey results. The survey results show that approaches to complex mixtures testing depend as much on which premises of toxicology (e.g., in vitro vs. dose-response studies) are adopted as on the methods selected for research.

The current EPA document presents many of the same conundrums as the previous attempts, particularly in its default positions. The current document implies that studies of complex field samples (such as Superfund site effluents) cannot yield generalizable data or conclusions because each site is unique. Although effluent composition may be quantitatively variable by chemical analyses, the toxicological profiles of these different mixtures may not be qualitatively distinctive. Degree of toxicological variation is an empirical question subject to testing. A related implication is that a slight (to be defined by the regulator) chemical difference negates the use of toxicological data from one mixture (e.g., site effluent) to assess a similar but non-congruent mixture (one in which the relative proportions of the constituents vary). Both current knowledge of toxicology, and the additive model itself, do not support such a position, which leads to the assertion that every complex mixture requires a full toxicological assessment--an impossible task.

The EPA is attempting to integrate several approaches to evaluating complex mixtures; this is a very important and lofty goal. However, there are few novel approaches in the document. The Committee is concerned that the risk evaluations are driven by carcinogenic potency, to the exclusion of other disease endpoints. More disturbingly, the proposed strategy document favors classical high-dose testing of single compounds as the components of an additive model.

The attempt to ascertain the efforts of other agencies and organizations is commendable. The NAS/NRC Committee in a similar survey, received answers corresponding to those received by EPA.

The USDA and the FDA have dealt with complex mixtures such as foods and their treatment by food additives; these efforts are given little attention in the EPA document. Such an oversight may indicate insufficient recognition that diet provides our most enduring exposure to complex mixtures; or it may signify too parochial a vision of the issue beyond EPA's literal responsibilities.

The cross-cutting section of the document is very helpful but must be expanded to integrate many more mixture scenarios.

In summary, the document is a useful attempt to integrate work by other government agencies and the broader scientific community. Its strength comes from its approach to the design of a research plan which, although vague and perhaps unrealistic, is nevertheless a prescription rather than another diagnosis. It offers premises that can be argued. Its weakness is a reliance on default assumptions that few would agree are scientifically defensible. In addition, the bibliography is incomplete; several workshops sponsored by non-governmental sources are missing from the reference list.

4. DISCUSSION AND CONCLUSIONS

Exposure to chemical mixtures, rather than to single chemicals, is far more common a problem in EPA's regulatory universe. The Agency has long recognized the issue and adopted interim procedures for calculating potential health risks posed by mixtures. In the absence of definitive information about the mixture itself, it assumes that the total risk of a mixture is equivalent to the sum of the risks associated with the mixture's constituents.

The scientific cogency of such an approach is debatable. Risk estimates based on simple additivity may either overestimate or underestimate the true risk. Underestimation is a particular concern of the Agency because of its public health responsibilities, and because of the data indicating that, under some circumstances, combinations of chemicals may be more toxic than would be predicted by summing the toxicity of the individual chemicals themselves.

To help narrow the gaps in knowledge about the health risks of mixtures, ORD, through HERL, has proposed a research strategy aimed primarily at what might be called a "synthesis logic," or bottom-up approach. Such a strategy presumes that once the constituents of a mixture have been identified, and the individual dose-response functions elaborated, the toxicity of the total blend can be calculated--provided that the mechanisms governing interactions are understood.

The Committee agrees that complex mixtures present a significant public health challenge and puzzle. We are not convinced, however, that ORD and HERL have devised the most useful research program for this purpose. As noted in various sections of the report, our major concerns are:

a. **Is the emphasis on "bottom-up" testing feasible?** Many complex mixtures contain dozens to thousands of individual constituents, including large numbers for which no toxicity data are available. Is it really feasible to predict the total potency of such a mixture by some combinatorial algorithm? Is that an appropriate alternative to testing mixture themselves? Should more scope be given to methods such as bioassay-directed fractionation? How will the "synthesis logic" be validated?

b. **How does the proposed research program plan to examine the variable of dose?** The great bulk of published experiments on mixtures consists of high-dose studies of binary combinations. EPA's risk analyses must be based on low environmental doses, whenever and however possible. With low incremental risks, as Committee members have noted, and as calculated in the NAS (1988) report, additive and multiplicative risks are indistinguishable, thereby eliminating the Agency's predominant concern about interactions exceeding the additivity assumption.

c. **What is the nature of the research to be carried out?** The Committee also remarked that, although the ORD/HERL proposal might be defended on toxicological grounds (that is, as consistent with toxicological practice), we

found it to be somewhat vague in its descriptions of specific research initiatives, lacking perspective on the public health implications, and unclear about the resources that would have to be committed to executing such a program.

In addition to requesting more specific plans for the research program, the Committee also asked for a fuller explanation of how the program related to the Reducing Risk framework destined to guide the Agency's future efforts and allocation of resources.

5. REFERENCES

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